At page 119, lines 16-28, please replace the existing paragraph with the following:

E3

(ii) Construction of S.erythraea ERM D1

Approximately 5-10 µg of pAVLD, isolated from E. coli TG1recO(pAVLD) was transformed into S. erythraea NRRL2338 and stable thiostrepton resistant colonies were isolated. One of these colonies was selected and total DNA was digested with PstI and analysed by Southern hybridisation employing as a probe the insert from plasmid pCRc which contains the fragment of the ery AI gene encoding the ketosynthase domain KS1. The analysis showed positively-hybridizing PstI fragments of 8.5 kbp, 4.8 kbp and 33 kbp, indicating the presence of two tandemly integrated copies of pAVLD.

SEQUENCE IDENTIFIER AMENDMENTS

In order to comply with the sequence listing rules, please amend the specification as follows to insert Sequence Identifiers where indicated:

E4

(Page 26, Line 6) The 1.6 kbp DNA segment encoding the loading domain of the erythromycin polyketide synthase from nucleotide 1 to 1680 was amplified by PCR employing the CloneAmp procedure (Raschtian, A. et al. Anal. Biochem. (1992) 91: 91-97) with the following two oligonucleotides as primers:

5'-ACGCGUACUAGUCCGATTAATTAAGGAGGACCATCATGGCGGACC TGTCAAAGCTC-3' (SEQ ID NO: 1) and

5'-AUGGAGAUCUCUCCGCTAGCGGTTCGCCGGGCGCCGCTTCGTTGGTCCGC GCGCGGGTTTCCC-3' (SEQ ID NO: 2) and using as template the DNA of plasmid pNTEP2. Approximately 30-60 ng of the PCR product (1.6 kbp) is digested with uracil DNA glycosylase for 30 minutes at 37°C in the presence of 25 ng of pAMP18 vector DNA (Gibco BRL), the mixture is cooled on ice and used to

Cont ¶4

transform E. coli TG1recO and individual colonies are checked for their plasmid content. The desired plasmid is identified by its restriction map and is designated pARLD

¶5

(Page 27, Line 14) pCJR101 (Figure 6) is a shuttle plasmid constructed to be used for expression of PKS genes in actinomycetes. It includes a ColEI replicon to allow it to replicate in E. coli, an SCP2* low copy number Streptomyces replicon (Bibb, M. J. and Hopwood, D. A. J. Gen. Microbiol. (1981) 126:427) and the actII-orf4 activator gene from the act cluster which activates transcription from the act promoter during the transition from growth phase to stationary phase in the vegetative mycelium. It is constructed as follows: an approximately 970 bp DNA fragment from pMF1015 (containing the actII-orf4 activator gene) (Fernandez-Moreno, M. A. et al. Cell (1991) 66:769-780) is amplified by PCR, using as primers the synthetic oligonucleotides: 5'-ACT AGT CCA CTG CCT CTC GGT AAA ATC CAG C-3' (SEQ ID NO: 3) and 5'-CTT AAG AGG GGC TCC ACC GCG TTC ACG GAC-3' (SEQ ID NO: 4), which also introduces flanking SpeI and AflII restriction sites. This fragment is introduced into the end-repaired AatII site of plasmid pUC19 to yield plasmid p18.14 (renamed pCJR18). An approximately 215 bp DNA fragment is amplified from pMV400 which contains the bidirectional promoter pair PactIII/PactI) (Parro, V. et al. Nucl. Acids Res. (1991) 19:2623-2627), using as primers the synthetic oligonucleotides 5'-ACA TTC TCT ACG CCT AAG TGT TCC CCT CCC TGC CTC-3' (SEQ ID NO: 5) and 5'-GTG ATG TAT GCT CAT ATG TGT CCT CCT TAA TTA ATC GAT GCG TTC GTC CGG TG-3' (SEQ ID NO: 6), which also introduces flanking NdeI and AflII sites. The PCR product is digested with NdeI and AflII and ligated with the plasmid p18.14 (pCJR18) previously cut with NdeI and AflII, to generate plasmid p19.4 (renamed pCJR19). A 1.1 kbp HindIII-SphI fragment containing the tsr gene, which confers resistance to thiostrepton, is obtained by PCR from plasmid pIJ922 (Lydiate, D. J. et al. Gene (1985) 35:223-235)

Cont E5

as template, using as primers the oligonucleotides 5'-TGA ACA CCA AGC TTG CCA GAG AGC GAC GAC TTC CCC-3' (SEQ ID NO: 7) and 5'-GAC AGA TTG CAT GCC CTT CGA GGA GTG CCC GCC CGG-3' (SEQ ID NO: 8) which also introduces flanking HindIII and SphI sites. The PCR product is digested with HindIII and SphI and ligated with plasmid p19.4 (pCJR19) cut with HindIII and SphI to obtain plasmid p20.5 (pCJR24). The plasmid pIJ922 is digested with BamHI and SstI and the fragment containing a portion of the fertility locus and the origin of replication (Lydiate, D. J. et al. Gene (1985) 35:223-235) is ligated into pUC19 digested with BamHI and Sst I to generate the bifunctional plasmid p16/2.2 (renamed pCJR16) (14.7 kbp). Plasmid p20.5 (pCJR24) is digested with SalI and SphI, the two larger fragments from the digest are purified by gel electrophoresis, and combined in a four-component ligation with plasmid 16/2.2 (pCJR16) which has been digested with XhoI and SphI. The ligation mixture is used to transform Streptomyces lividans and colonies are selected in the presence of thiostrepton. One such colony is shown to contain the desired plasmid pCJR101 (approx. 12.4 kbp), identified by its restriction pattern.

(Page 28, Line 37) The construction of plasmid pCJR29 (pCJR110) is illustrated in Figure 7. A 1.1 kbp HindIII-XhoI fragment containing the tsr gene, which confers resistance to thiostrepton, is obtained by PCR from plasmid pIJ922 as template, using as primers the oligonucleotides 5'-TGA ACA CCA AGC TTG CCA GAG AGC GAC GAC TTC CCC-3' (SEQ ID NO: 7) and 5'-GAC AGA TTC TCG AGC CTT CGA GGA GTG CCC GCC CGG-3' (SEQ ID NO: 9) which also introduces flanking HindIII and XhoI sites. The PCR product is digested with HindIII and XhoI and ligated with plasmid 16/2.2 (pCJR16) which has been digested with HindIII and XhoI, to generate plasmid 22.1 (pCJR25). Plasmid p22.1 (pCJR25) is digested with HindIII and SphI and ligated with plasmid p19.4 (pCJR19) which has been digested with

E6

HindIII and SphI, to produce the desired plasmid pCJR29 (pCJR110) (approx. 12.4 kbp), identified by its restriction pattern. Plasmid pCJR29 (pCJR110) differs from pCJR101 in the orientation of the tsr gene, the actII-orf4 gene and the actI/actIII promoter, with respect to the SCP2*-derived origin of replication.

(Page 29, Line 22) Plasmid pRM52 is a derivative of plasmid pRM5 (McDaniel, R. et al. Science, (1993) 262:1546-1550). pRM5 was first linearised by digestion with NdeI, end-repaired and then religated to produce pRM51. pRM51 was cut with PacI and NsiI and the large PacI-NsiI fragment was isolated and ligated to a short double-stranded oligonucleotide linker containing an NdeI site and constructed from the synthetic oligonucleotides 5'-TAAGGAGGACACATATGCA-3' (SEQ ID NO: 10) and 5'-TAATTCCTCCTGTGTAT-3' (SEQ ID NO: 11) which were annealed together. The ligation mixture was transformed into E. coli TGIrecO and isolated colonies were screened for their plasmid content. The desired plasmid (19.6 kbp) was identified by its restriction map and was designated pRM52.

(Page 30, Line 25) A 4.0 kb KpnI fragment extending from 1.4 kbp upstream of the correct eryAI start codon as previously determined (Caffrey, P. et al. FEBS Letters (1992) 304:225-228), to 2.6 kbp inside the eryAI gene of S. erythraea, was excised from plasmid pBK25 (Bevitt, D. J. et al. Eur. J. Biochem. (1992) 204:39-49) and cloned into pUC18 to obtain plasmid pBK6.12. DNA of this plasmid was used as the template for a PCR reaction to obtain a 360 bp product in which a unique Nde I site is created at the start codon of eryAI and a unique SmaI site is created at the other end of the PCR product. The oligonucleotides used were 5'-CCC ATA TGG CGG ACC TGT CAA AGC-3' (SEQ ID NO: 12) and 5'-ATT GCG CGC CCT GGC CCG GGA A-3' (SEQ ID NO: 13). The product was end-

Cont E8

repaired and ligated into SmaI cut pUC18, and transformed into E. coli TG1recO.

E9

(Page 32, Line 3) A ClaI-EcoRI polylinker, bearing unique restriction sites for XbaI and for HindIII was constructed, from the following complementary synthetic oligonucleotides: 5'-AATTCATAGTCTAGAAGCTTAT-3' (SEQ ID NO: 14) and 5'-CGATAAGCTTCTAGACTATG-3' (SEQ ID NO: 15) The polylinker was ligated into plasmid pNTE5, which had been digested with ClaI and EcoRI to remove a 2.3 kbp ClaI-EcoRI fragment. The ligation mixture was used to transform E. coli TG1recO and individual colonies were screened for their plasmid content. One plasmid containing the polylinker was identified and designated pNTEP2.

E10

(Page 35, Line 6) Plasmid pCRabc (Figure 10) was constructed as follows. Three separate PCR reactions were conducted: First, 20 pmol each of synthetic oligonucleotides A1 (5'-CTC GTC GGT GGC TTT GCG-3'; SEQ ID NO: 16) and A2 (5'-CCC GGG AAA AAC GAA GAC TAG TGG CGC GGA CGG CCG-3'; SEQ ID NO: 17) were used to amplify a 1.0 kbp product from 100 ng pNCO12 template. The PCR product was end-repaired, phosphorylated and cloned into SmaI-cut pUC18 to obtain plasmid pCRa. Secondly, 20 pmol each of synthetic oligonucleotides C1 (5'-CAC GCG CAG CGC GGC GGA-3'; SEQ ID NO: 18) and C2 (5'-CGAA CCG CTA GCG GTC GTC GCG ATG GCC T-3'; SEQ ID NO: 19) were used to amplify a 1.5 kbp product from 100 ng pNC012 template. The product was end-repaired, phosphorylated and cloned into SmaI-cut pUC18 to obtain plasmid pCRc. Thirdly, 20 pmol each of synthetic oligonucleotides B1 (5'-GTGGCCCGGCCGTCCGCGCCACTAGTCTTCGTTTTT-3'; SEQ ID NO: 20) and B2 (5'-AACAGCTAGCGGTTCGTCCGCCGCTGCCGTGCC-3'; SEQ ID NO: 21) were used to amplify a 1.4 kbp product from 100 ng pVE3.4 template. The product was end-repaired, phosphorylated and cloned into SmaI-cut pUC18 to obtain plasmid pCRb.

E11

(Page 42, Line 7) Plasmid pNEWAVETE was digested with EcoRI and HindIII and the vector was purified by gel electrophoresis. A synthetic oligonucleotide double-stranded insert encoding a 6-histidine tag and possessing these sites at either end (shown below) was ligated to the vector.

(5'-AATTCACATCACCATCACCATCACTAGTAGGAGGTCTGGCCATCTAGA-3'; SEQ ID NO: 22)
(3'-GTAGTGGTAGTGGTAGTGATCATCCTCCAGACCGGTAGATCTTCGC-5'; SEQ ID: 23)

E12

(Page 44, Line 19) For the PCR amplification of DNA for module 1, the following synthetic oligonucleotides were used as mutagenic primers, one containing an EcoRV site and the other a BglII site: 5'-GCAGGGATATCGCACGTTCCTGG-3' (SEQ ID NO: 24) and 5'-CGCCGAGATCTGCGAAGGCCTGGTCGGCGGG-3' (SEQ ID NO: 25)

E13

(Page 44, Line 37) For PCR amplification of the DNA for the 5' end of module 2 and the thioesterase domain, the following oligonucleotides containing respectively a Bgl II site and an EcoRI site, were used as mutagenic primers:

5'-ATGAATTCCCTCCGCCCAGCCAG-3' (SEQ ID NO: 26)
and 5'-ACAGATCTCGGCTTCGACTCGCTGACCG-3' (SEQ ID NO: 27)

E14

(Page 48, Line 1) The 4.7 kbp DNA segment of the rapC gene encoding module 12 of the rapamycin PKS was amplified by PCR employing the CloneAmp procedure (Raschtian, A. et al. Anal. Biochem. (1992) 91:91-97) and with the following two oligonucleotides as primers:
5'-ACGCGUACUAGUCAGATCTGGGCATCAATTCGCTGACCGCGGTGGAACTGCGCAA-3' (SEQ ID NO: 28)
and 5'-AUGGAGAUCUCUCAGATCTTGAATGCGGCGGCTGCGGGGATGGTGCTGGCGTCA-3' (SEQ ID NO: 29), and using as template the DNA of clone_λ-1C (Schwecke, T. et al. Proc. Natl. Acad. Sci. USA (1995)

Cont E14

92:7839-7843). Approximately 30-60 ng of the PCR product (4.7 kbp) is digested with uracil DNA glycosylase for 30 minutes at 37°C in the presence of 25 ng pAMP18 vector DNA (Gibco BRL), the mixture is cooled on ice and transformed into E. coli TG1recO and individual colonies are checked for their plasmid content. The desired plasmid (7.4 kbp) is identified by its restriction map and is designated pARRAP.

E15

(Page 49, Line 3) Plasmid pAR32 contains an insert that can be excised by digestion with NdeI and XbaI, but there is an additional NdeI site in the insert that must be specifically protected against cleavage. This is done using the RecA protection method (Koob, M. et al. Nucl. Acids Res. (1992) 20:5831-5835)). The synthetic oligonucleotide 5'-GCACCCACGACGCCACCACCACATATGCCCTGCACCCTGCCCTCC-3' (SEQ ID NO: 30) (in which the NdeI site is underlined) is used together with purified RecA protein and ATP_S, to form a stable triplex DNA-protein complex that specifically protects the internal NdeI site in rap module 12 from digestion. The protected plasmid pAR32 is digested with NdeI and XbaI, producing the desired full-length insert (13.1 kbp), and this is ligated with plasmid pRM52 (Example 4) which has been digested with NdeI and XbaI. The ligation mixture is transformed into E. coli TG1 recO and individual colonies are screened for their plasmid content. The desired plasmid pAR33 is identified by its restriction pattern.

E16

(Page 50, Line 34) The segment of the ery AI gene from nucleotide 1 to nucleotide 1673, encoding the loading AT-ACP didomain, was amplified by PCR employing the CloneAmp procedure with the following two oligodeoxynucleotides as primers:
5'-ACGCGUACUAGUCCGATTAATTAAGGAGGACCATCAATGGCGGACCTGTCAAAGCTC-3' (SEQ ID NO: 31) and 5'-AUGGAGAUCUCUCCGCTAGCGGTTCGCCGGGCGCC GCTTCGTTGGTCCGCGCGCGGGTTTCCC-3' (SEQ ID NO: 2)and plasmid

Cont E16 pBK6.12 (Example 5) as template, to give ~~plasmid pARLD.~~

E17 (Page 51, Line 15) The segment of the rapC gene of S. hygroscopicus (Schwecke, T. et al. Proc. Natl. Acad. Sci. USA (1995) 92:7839-7843) from nucleotide 112 to nucleotide 2095, the 5'- end of the DNA encoding rap module 11, is amplified by PCR employing the CloneAmp procedure with the following two oligodeoxynucleotides as primers:

5'-AUGGAGAUCUCUCCGCTAGCGATTGTGGGTATGGCG-3' (SEQ ID NO: 32) and
5'-ACGCGUACUAGUCCATGCATCTGCAGCACGGCGGCCTCATCACCGGA-3' (SEQ ID NO: 33)
and the DNA of recombinant bacteriophage _λ-1C (Schwecke, T. et al., Proc. Natl. Acad. Sci. USA (1995) 92:7839-7843) as the template. Approximately 30-60 ng of the PCR product (2.0 kbp) is digested with uracil DNA glycosylase for 30 min at 37°C in the presence of 25 ng pAMP18 vector DNA, the mixture is cooled on ice and transformed into E. coli TG1 recO and individual colonies checked for their plasmid content. The desired plasmid (4.7 kbp) is identified by its restriction map and is ~~designated pAR11.~~

E18 (Page 52, Line 1) The segment of the rapC gene of S. hygroscopicus (Schwecke, T. et al., Proc. Natl. Acad. Sci. USA (1995) 92:7839-7843) from nucleotide 7405 to nucleotide 9396, the 3' end of the DNA encoding rap module 12, is amplified by PCR employing the CloneAmp procedure with the following two oligodeoxynucleotides as primers:

5'-ACGCGUACUAGUCCATGCATTCCCGGAGCGGCGATCTGTGG-3' (SEQ ID NO: 34) and 5'-AUGGAGAUCUCUCCCGCGGCCGCGCTGTC ACGCACCAGCTTCAGCAGTGCGTC-3' (SEQ ID NO: 35) and the DNA of recombinant bacteriophage λ-1C (Schwecke, T. et al., Proc. Natl. Acad. Sci. USA (1995) 92:7839-7843) as template.

Cont E18

Approximately 30-60 ng of the PCR product (2.0 kbp) is digested with uracil DNA glycosylase for 30 minutes at 37°C in the presence of 25 ng pAMP18 vector DNA, the mixture is cooled on ice and tranformed into E. coli TG1recO and individual colonies are checked for their plasmid content. The desired plasmid (4.7 kbp) is identified by its restriction map and is designated pAR12.

E19

(Page 52, Line 24) The 1.3 kbp segment of the eryAIII gene, extending by 132 nucleotides 3' of the eryAIII stop codon to a KpnI site, and encoding the C-terminal chain-terminating thioesterase/cyclase of DEBS, is amplified by PCR employing the CloneAmp procedure with the following two oligodeoxynucleotides as primers: 5'-ACGCGUACUAGUCCGCGGCCGCGATCCTCGGGCATTCCAGC-3' (SEQ ID NO: 36) and 5'-AUGGAGAUCUCUAAGCATTGGTAACTGTC-3' (SEQ ID NO: 37), and plasmid pEXDB3 (Roberts, G. A. et al. Eur J. Biochem. (1993) 214:305-311) as the template. Approximately 30-60 ng of the PCR product (1.3 kbp) is digested with uracil DNA glycosylase for 30 min at 37°C in the presence of 25 ng pAMP18 vector DNA, the mixture is cooled on ice and transformed into E. coli TG1 recO and individual colonies checked for their plasmid content. The desired plasmid (4.0 kbp) is identified by its restriction map and is designated pARTE.

E20

(Page 53, Line 8) The 1.3 kbp segment of plasmid pBR322 containing the tetracycline resistance gene is amplified by the CloneAmp procedure with the following two oligodeoxynucleotides as primers:

5'-ACGCGUACUAGUATCTAGACCATGCATGTTTGACAGCTTATCATC-3' (SEQ ID NO: 38) and 5'-AUGGAGAUCUCUATCTAGACCATGCATGCCGCCGGCTTCCATTCA-3' (SEQ ID NO: 39) and plasmid pBR322 as the template. Approximately 30-60 ng of the PCR product (1.3 kbp) is

Cont E20 digested with uracil DNA glycosylase for 30 minutes at 37°C in the presence of 25 ng pAMP18 vector DNA, the mixture is cooled on ice and transformed into E. coli TG1recO and individual colonies are checked for their plasmid content. The desired plasmid (4.0 kbp) is identified by its restriction map and is designated pARTr.

E21 (Page 58, Line 15) 50 pmol of each of synthetic oligonucleotides 8985 (5'-GAGCAGTCGTTCCGAGATCTCGGCTTCGATTCA-3'; SEQ ID NO: 40) which introduced a BglII site and 9204 (5'-GGGAGGAGATCAGATCCCAGAAGT-3'; SEQ ID NO: 41) were used by PCR to amplify a 300 bp product from 60 ng pIG70ΔEco. The PCR product was end-repaired, phosphorylated and ligated into pUC18 that had been linearised with SmaI and dephosphorylated. The ligation mixture was used to transform E. coli TG1 recO and individual colonies were checked for their plasmid content. The orientation of pIGPCRstart was identified by a double restriction enzyme digest with EcoRI and BglII to give a pattern that included a 300 bp fragment.

E22 (Page 58, Line 30) 50 pmol of each of synthetic oligonucleotides 8986 (5'-GAGGGAGTCGAACCGAGATCTCGGAACGCGCGG-3'; SEQ ID NO: 42) which introduced a BglII site and 9205 (5'-GGGGGATCCTGGGGTCGGCCGGGCAGGGCAA-3'; SEQ ID NO: 43) were used by PCR to amplify a 440 bp product from 60 ng pIG71ΔSac. The PCR product was end-repaired, phosphorylated and ligated into pUC18 that had been linearised with SmaI and dephosphorylated. The ligation mixture was used to transform E. coli TG1 recO and individual colonies were checked for their plasmid content. The orientation of pIGPCRend was identified by its restriction enzyme digest pattern.

E23 (Page 61, Line 7) For the PCR amplification for plasmid pMO07, the following synthetic oligonucleotides

Cont E23 were used as mutagenic primers, one containing a HindIII site and the other an EcoRV site:
5' -GTCTCAAGCTTCGGCATCAGCGGCACCAA- 3' (SEQ ID NO: 44)
and 5' -CGTGCGATATCCCTGCTCGGCGAGCGCA-3' (SEQ ID NO: 45)

E24 (Page 61, Line 13) For the PCR amplification for plasmid pMO08, the following synthetic oligonucleotides were used as mutagenic primers, one containing a PstI site and the other a HindIII site:
5' -CATGGCCTGCAGGCTGCCCGGGGAGGTCGACT- 3' (SEQ ID NO: 46)
and 5' -CCCGAAGCTTGACACACCTGCCCGGCGCACCCCGT- 3' (SEQ ID NO: 47)

E25 (Page 61, Line 20) For the PCR amplification for plasmid pMO09, the following synthetic oligonucleotides were used as mutagenic primers, one containing a MunI site and the other a PstI site: 5' -
GCGCGCCAATTGCGTGCACATCTCGAT- 3' (SEQ ID NO: 48)
and 5' -CCTGCAGGCCATCGCGACGACCGCGACCGGTTCGCCG- 3' (SEQ ID NO: 49)

E26 (Page 63, Line 7) For the PCR amplification for plasmid pKSA, the following synthetic oligonucleotides were used as mutagenic primers, one containing a PstI site and the other a HindIII site: 5' -GATGGCCTGCAGGCTGCCCGGCGGTGTGAGCA- 3' (SEQ ID NO: 50) For the PCR amplification for plasmid pKSA, the following synthetic oligonucleotides were used as mutagenic primers, one containing a PstI site and the other a HindIII site: 5' -GATGGCCTGCAGGCTGCCCGGCGGTGTGAGCA- 3' (SEQ ID NO: 50)
and 5' -GCCGAAGCTTGAGACCCCCGCCCGGCGCGGTCGC- 3' (SEQ ID NO: 51)

E27 (Page 63, Line 14) For the PCR amplification for plasmid pKSB, the following synthetic oligonucleotides were used as mutagenic primers, one containing an EspI site and the other a PstI site:

Sub F

Look E26 E27

5' -TGGCTTCGCTGGCGGACACGCTCAG- 3' (SEQ ID NO: 52)
and 5' -CCTGCAGGCCATGCCGACGATCGCGATCGGCT- 3' (SEQ ID NO: 53)

E28

(Page 63, Line 21) For the PCR amplification for plasmid pKSC, the following synthetic oligonucleotides were used as mutagenic primers, one containing a HindIII site and the other a BspEI site:
5' -GTCAAGCTTCGGGGTGAGCGGGACGAA- 3' (SEQ ID NO: 54)
and 5' -GCGTCCGGACGTGGCTCCAGCA-3' (SEQ ID NO: 55)

E ~~28~~ 29

(Page 73, Line 6) The approximately 0.46 kbp DNA fragment of the eryAI gene of S. erythraea was amplified by PCR using as primers the synthetic oligonucleotides: 5'-GGAGTACTGCGAGGGCGTGGGCAT-3' (SEQ ID NO: 56) and 5'-CACCTAGGACCGCTTCCCAGTCGACC-3'(SEQ ID NO: 57) and plasmid pNTEPH as template. The PCR product was treated with T4 polynucleotide kinase and then ligated with plasmid pUC18, which had been linearised by digestion with SmaI and then treated with alkaline phosphatase. The ligation mixture was used to transform E. coli DH10B and individual colonies were checked for their plasmid content. The desired plasmid pJLK01 was identified by its restriction pattern and DNA sequencing.

E30

(Page 73, Line 22) The approximately 1.47 kbp DNA fragment of the eryAI gene of S. erythraea was amplified by PCR using as primers the synthetic oligonucleotides: 5'-TACCTAGGCCGGGCCGGACTGGTCGACCTGCCGGGTT-3' (SEQ ID NO: 58) and 5'-ATCCTCAGGCTCTCCGTCTCCGGTTCTCC-3' (SEQ ID NO: 59) and plasmid pNTEPH as template. The PCR product was treated with T4 polynucleotide kinase and then ligated with plasmid pUC18, which had been linearised by digestion with SmaI and then treated with alkaline phosphatase. The ligation mixture was used to transform E. coli DH10B and individual colonies were checked for their plasmid content. The desired plasmid pJLK08 was identified by its restriction pattern and DNA sequencing.

(Page 74, Line 1) The approximately 1.12 kbp DNA fragment of the eryAI gene of S. erythraea was amplified by PCR using as primers the synthetic oligonucleotides: 5'-TACCTGAGGGACCGGCTAGCGGGTCTGCCGCGTG-3'(SEQ ID NO: 60) and 5'-CTTCTAGACTATGAATTCCCTCCGCCCAGC-3' (SEQ ID NO: 61) and plasmid pNTEPH as template. The PCR product was treated with T4 polynucleotide kinase and then ligated with plasmid pUC18, which had been linearised by digestion with SmaI and then treated with alkaline phosphatase. The ligation mixture was used to transform E. coli DH10B and individual colonies were checked for their plasmid content. The desired plasmid pJLK09 was identified by its restriction pattern and DNA sequencing.

(Page 75, Line 22) The approximately 3.3 kbp DNA of the rapC gene of S. hygroscopicus encoding the reduction loop of module 13 was amplified by PCR using as primers the synthetic oligonucleotides: 5'-CGCCTAGGCACCACCACAACCCGGGTACTGGACC-3'(SEQ ID NO: 62) and 5'-TAGCTAGCCGGGCGCTCAGGGGCTGCGAGCCGACCT-3' (SEQ ID NO: 63) and cosmid cos 31 (Schwecke, T. et al. (1995) Proc. Natl. Acad. Sci. USA 92:7839-7843) as template. The PCR product was treated with T4 polynucleotide kinase and then ligated with plasmid pUC18, which had been linearised by digestion with SmaI and then treated with alkaline phosphatase. The ligation mixture was used to transform E. coli DH10B and individual colonies were checked for their plasmid content. The desired plasmid pJLK14 was identified by its restriction pattern and DNA sequencing.

(Page 77, Line 17) The approximately 2.8 kbp DNA fragment of the rapA gene of S. hygroscopicus encoding the reduction loop of module 4 was amplified by PCR using as primers the synthetic oligonucleotides: 5'-CCTAGGCACCACCACGGCCCGGGTGCTGGACCTT -3' (SEQ ID NO: 64) and 5'-CCTCAGGCTGTCACCGGTAGAGGCGGCCCT-3' (SEQ ID NO: 65) and cosmid cos 25 (Schwecke, T. et al. (1995) Proc. Natl. Acad.

Cont E33

Sci. USA 92:7839-7843) as template. The PCR product was treated with T4 polynucleotide kinase and then ligated with plasmid pUC18, which had been linearised by digestion with SmaI and then treated with alkaline phosphatase. The ligation mixture was used to transform E. coli DH10B and individual colonies were checked for their plasmid content. The desired plasmid pJLK16 was identified by its restriction pattern and DNA sequencing.

E34

(Page 79, Line 14) For the PCR amplification of an approximately 1.3 kbp DNA fragment for plasmid pJLK19, the following synthetic oligonucleotides were used as primers:
5' -GTCAAGCTTCGGGGTGAGCGGGACGAA- 3'(SEQ ID NO: 54) and 5' -ATCCTAGGACCGCTTCCCAGTCGACCGCGACA- 3' SEQ ID NO: 66) PCR was carried out on pNTEPH as template. The PCR product was treated with T4 polynucleotide kinase and then ligated with plasmid pUC18, which had been linearised by digestion with SmaI and then treated with alkaline phosphatase. The ligation mixture was used to tranform E. coli DH10B and individual colonies were checked for their plasmid content. The desired plasmid pJKL19 was identified by its restriction pattern.

E35

(Page 82, Line 19) The 1.4 kbp segment of plasmid pNTEP2 containing from nucleotide 9838 to 11214 (encoding amino acids 3279 to the end of DEBS1-TE) is amplified by PCR with the following two synthetic oligonucleotides as primers

5'-GCCACTAGTGTGGCGTGGGGGCTGTGGG-3' (SEQ ID NO: 67) and 5'-TGAATTCCCTCCGCCCAGCCAGGCGTCGAT-3' (SEQ ID NO: 68) and plasmid pNTEP2 as template. The PCR product was end-repaired and ligated with plasmid pUC18, which had been linearised by digestion with SmaI and then treated

Cont E35

with alkaline phosphatase. The ligation mixture was used to transform E. coli DH10B and individual colonies were checked for their plasmid content. The desired plasmid p37 in which an SpeI site was introduced at the 5' end of this fragment was identified by its restriction pattern and by DNA sequencing.

E36

(Page 83, line 10) The 1.1 kbp DNA segment of the eryAI gene of S. erythraea extending from nucleotide 8202 to nucleotide 9306 was amplified by PCR using as primers the synthetic oligonucleotides:
5'-CCTGGAGTACTGCGAGGGCGTG-3' (SEQ ID NO: 69) and
5'-CTGACTAGTGGCGGTGACGTGGGCGGGGGAAA-3' (SEQ ID NO: 70) and plasmid pNTEP2 as template. The PCR product was end-repaired and ligated with plasmid pUC18, which had been linearised by digestion with SmaI and then treated with alkaline phosphatase. The ligation mixture was used to transform E. coli DH10B and individual colonies were checked for their plasmid content. The desired plasmid pSCA7 in which an SpeI site has been introduced at the 3' end of this PCR product was identified by its restriction pattern and by DNA sequencing.

E37

(Page 85, Line 27) The 1.9 kbp segment of pUC1-0 from nucleotide 8715 to 10645 was amplified by PCR using as primers the synthetic oligonucleotides:
5'-CCCCTGCAGCCGGACCGCACCACCCCTCGTGACGA-3' (SEQ ID NO: 71) and
5'-CTTCTAGACTATGAATTCCCTCCGCCCAGC (SEQ ID NO: 61) and the DNA of pUC1-0 as template. The PCR product was end repaired and ligated with plasmid pUC18, which had been linearised by digestion with SmaI and treated with alkaline phosphatase. The ligation mixture was used to transform E. coli DH10B and individual colonies were checked for their plasmid content. The desired plasmid designated p1-0 was identified by restriction analysis and DNA sequencing.

E38

(Page 86, Line 5) The 60bp segment of eryAIII from nucleotide 7006 to 7066 was amplified by PCR using as primers the synthetic oligonucleotides:

5'-GGCGGAACGTCTTCCCGGCGGCACCT-3' (SEQ ID NO: 72) and
5'-CCCCTGCAGCCAGTACCGCTGGGGCTCGAA-3' (SEQ ID NO: 73) and
pEXDB3 (Roberts, G. A., et al. (1993) Eur. J. Biochem. 214:305-311) as template. The PCR product was end-repaired and ligated with plasmid pUC18, which had been linearised by digestion with SmaI and then treated with alkaline phosphatase. The ligation mixture was used to transform E. coli DH10B and individual colonies were checked for their plasmid content. The desired plasmid designated pD3P was identified by restriction analysis and DNA sequencing.

E39

(Page 90, Line 18) The approximately 1.3 kbp DNA segment of the eryAI gene of S. erythraea extending from nucleotide 1948 to nucleotide 3273 of eryAI (Donadio, S. et al. Science (1991) 252:675-679) was amplified by PCR employing as primers the synthetic oligonucleotides:
5'-CATGCTCGAGCTCTCCTGGGAAGT-3' (SEQ ID NO: 74) and
5'-CAACCCTGGCCAGGGAAGACGAAGACGG-3' (SEQ ID NO: 75), and plasmid pNTEP2 (Example 5) as template. The PCR product was end-repaired and ligated with plasmid pUC18, which had been linearised by digestion with SmaI and then treated with alkaline phosphatase. The ligation mixture was used to transform E. coli TG1 recO and individual colonies were checked for their plasmid content. The desired plasmid pMO1 (3.9 kbp), in which the StuI site bordering the insert is adjacent to the HindIII site in the polylinker, was identified by its restriction pattern.

E39
40

(Page 91, Line 1) The approximately 0.85 kbp DNA segment of the rapA gene of S. hygroscopicus, extending from nucleotide 1643 to nucleotide 2486 of rapA, was amplified by

PCR employing as primers the following oligonucleotides:

5'-TTCCCTGGCCAGGGGTCGCAGCGTG-3' (SEQ ID NO: 76) and
5'-CACCTAGGACCGCGGACCACTCGAC-3' (SEQ ID NO: 77), and the DNA from the recombinant bacteriophage _-1E (Schwecke, T. et al. Proc. Natl. Acad. Sci. USA (1995) 92:7839-7843) as the template. The PCR product was end-repaired and ligated with plasmid pUC18, which had been linearised by digestion with SmaI and then treated with alkaline phosphatase. The ligation mixture was used to transform E. coli TG1 recO and individual colonies were checked for their plasmid content. The desired plasmid pMO2 (3.5 kbp) was identified by its restriction pattern.

E40 41 (Page 91, Line 20) The approximately 1.7 kbp DNA segment of the eryAI gene of S. erythraea extending from nucleotide 4128 to nucleotide 5928 of eryAI, was amplified by PCR employing as primers the synthetic oligonucleotides:
5'-TGGCCAGGGAGTCGGTGCACCTAGGCA-3' (SEQ ID NO: 78) and
5'-GCCGACAGCGAGTCGACGCCGAGTT-3' (SEQ ID NO: 79) and plasmid pNTEP2 as template. The PCR product was end-repaired and ligated with plasmid pUC18, which had been linearised by digestion with SmaI and then treated with alkaline phosphatase. The ligation mixture was used to transform E. coli TG1 recO and individual colonies were checked for their plasmid content. The desired plasmid pMO3 (4.4 kbp), in which the BalI and AvrII sites are adjacent to the HindIII site of the polylinker, was identified by its restriction pattern.

E42 (Page 100, Line 15) The approximately 0.9 kbp DNA segment of the ATX domain was amplified by PCR employing as primers the following oligonucleotides:
5' CTGGCCAGGGCGCGCAATGGCCGAGCAT 3' (SEQ ID NO: 80) and
5' CCCTAGGAGTCGCCGGCAGTCCAGCGCGGCGCCC 3' (SEQ ID NO: 81) using the DNA from the cosmid pSCIN02 as the template. The PCR

Cont
E42

product was end-repaired and ligated with plasmid pUC18, which had been linearised by digestion with SmaI and then treated with alkaline phosphatase. The ligation mixture was used to transform E. coli TG1 recO and individual colonies were checked for their plasmid content. The desired plasmid pMO33 (3.5 kbp) was identified by its restriction pattern.

E43

(Page 100, Line 30) Plasmid pMO34 is a derivative of pMO6 with a polycloning site inserted after the stop codon of the inserted D1-AT2 gene. Plasmid pMO6 was digested wih EcoRI and HindIII and annealed with two oligonucleotides forming the double-stranded region of the polycloning site:
5' AATTCATAACTAGTAGGAGGTCTGGCCATCTAGA 3'(SEQ ID NO: 82)
and 5' TCGAAGATCTACCGGTCTGGAGGATGATCAATAC 3'(SEQ ID NO: 83).
The mixture was ligated and transformed into E. coli TG1 recO. Individual colonies were checked for their plasmid content. The desired plasmid pMO34 (13.5 kbp) was identified by its restriction pattern.

E44

(Page 104, Line 16) The approximately 1.0 kbp DNA segment of the eryAI gene of S.erythraea extending from nucleotide 6696 to nucleotide 7707 of eryAI (Donadio. S. et al., Science (1991) 252, 675-679) was amplified by PCR employing as primers synthetic oligonucleotides:
5' GGCGGGTCCGGAGGTGTTCACCGAGTT 3'(SEQ ID NO: 84)
and 5' ACCTTGGCCAGGGAAGACGAACACTGA 3' (SEQ ID NO: 85), and plasmid pNTEp2 as a template. The PCR product was end-repaired and ligated with plasmid pUC18, which had been linearised by digestion with SmaI and then treated with alkaline phosphatase. The ligation mixture was used to transform E.coli TG1 recO and individual colonies were checked for their plasmid content. The desired plasmid pMO25 (3.6 kbp), in which the StuI site bordering the insert is adjacent to the HindIII site in the polylinker, was identified by its restriction pattern.

E45

(Page 105, Line 13) The approximately 0.6 kbp DNA segment of the eryAI gene of S. erythraea extending from nucleotlde 8660 to nucleotide 9258 of eryAI, was amplified by PCR employing as primers the synthetic oligonucleotides:
5' TCCTAGGCCGGGCCGGACTGGTCGACCTGCCGGGTT 3' (SEQ ID NO: 86)
and 5' AAACACCGCGACCTGGTCCTCCGAGC 3' (SEQ ID NO: 87), and plasmid pNTEP2 as template. The PCR product was end-repaired and ligated with plasmid pUC18, which had been linearised by digestion with SmaI and then treated with aIkaline phosphatase. The ligation mixture was used to transform E. coli TG1 recO and individual colonies were checked for their plasmid content. The desired plasmid pMO26 (3.2 kbp), in which the AvrII site is adjacent to the HindIII site of the polylinker, was identified by its restriction pattern.

E46

(Page 112, Line 5) The 250 bp DNA segment of the eryAIII gene of S. erythraea extending from nucleotide 4807 to nucleotide 5052 of eryAIII, was amplified by PCR employing as primers the synthetic oligonucleotides:
5' TTTGCTAGCGATCGTCGGCATGGCGTGCCGGTT3'(SEQ ID NO: 88)
5'CCCACGAGATCTCCAGCATGATCC3' (SEQ ID NO: 89)
The plasmid pEXD3 was used as a template. The PCR product was end-repaired and ligated with pUC18, which had been linearised by digestion with SmaI and then treated with alkaline phosphatase. The ligation mixture was used to transform E.Coli TG1 recO and individual colonies were checked for their plasmid content. The desired plasmid pCAR5 in which the NheI site is adjacent to the EcoRI site of the polylinker, was identified by its restriction pattern and sequence analysis.

E47

(Page 117, Line 1) The 450 bp DNA segment of the eryAI gene of S.erythraea extending from nucleotide 1 to nucleotide 10631 of eryAI, was amplified by PCR employing as primers the synthetic oligonucleotides:(bases in bold letters denote the restriction enzyme sites).